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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
09/555,574	10/23/2000	Jean-Paul Behr	0652.2090000	9271

7590 03/27/2002

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EXAMINER
SCHMIDT, MARY M

ART UNIT	PAPER NUMBER
1635	

DATE MAILED: 03/27/2002 14

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	09/555,574	BEHR ET AL.	
	Examiner	Art Unit	
	Mary Schmidt	1635	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *08 January 2002* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-48* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-48* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *23 October 2000* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

1. Please note that the Examiner of record for this Application has changed. Please direct all future correspondence to Examiner Schmidt. Please note the concluding remarks below for information on how to reach the Examiner.

2. Please note that the requirement for restriction made in the previous Official Action mailed 08/08/01 is withdrawn. The instant Application was filed under 35 U.S.C. 371 as a National Phase of PCT/EP98/07695. No lack of unity was found among the claims. **Claims 1-48 are pending** for consideration on the merits below. The instant Action is made non-final in view of the new consideration of all the outstanding claims. The 35 U.S.C. 102 rejections made in the previous Official Action mailed 08/08/01 were removed in view of the amendments to claims 1-48 and Applicants's response in the reply filed 01/08/02. Specifically, the claims were amended to recite that covalent bonds are formed upon condensation of the cationic precursor molecules with the nucleic acid molecules without crosslinking any of the nucleic acids to the precursor molecules which was not explicitly taught by the references cited in the prior Official Action.

Claim Rejections - 35 USC § 112

3. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to

make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

4. Claims 1-48 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for guanidyl-cysteine-decylamide ($C_{10}C^{G+}$) and ornithyl-cysteine-dodecylamide ($C_{12}CO$) and methods of making and using said compositions in cells in culture and rodents, does not reasonably provide enablement for the scope of compositions claimed nor methods of making and using said compositions in cells in any whole organism such as humans for therapeutic purposes. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention commensurate in scope with these claims.

Please note, the instant rejection is a modification of the 35 U.S.C. 112, scope of enablement rejections made in the Official Action mailed 08/08/01. The indicated allowability of claim 21 is withdrawn in view of the following arguments. The pertinent arguments made by Applicant in response to the previous 35 U.S.C. 112, scope of enablement, rejections are addressed following the instant rejection.

Claim **1** as amended is drawn to a particle for transfecting higher eukaryotic cells with nucleic acid molecules in vitro and in vivo comprising one or more nucleic acid molecules condensed by organic cationic molecules, said particle being obtained by (1) condensing said one or more nucleic acid molecules with identical or different organic cationic precursor molecules without crosslinking any of said one or more nucleic acid molecules, and (2) thereafter linking the precursor molecules to each other with one or more covalent bonds on the condensed one or

more nucleic acid molecules. Claim **2** specifies wherein the cationic molecules are lipids obtained by dimerization or oligomerization of cationic detergent precursor molecules. Claim **3** specifies wherein the cationic detergent precursor molecules comprise (a) at least one functional group for binding to one or more other detergent molecules, (b) at least one lipophilic residue, c) a non-toxic recipient backbone, (d) a cationic group for binding to nucleic acid molecules. Claim **4** specifies wherein the functional group of the cationic precursor detergent molecules for binding to other detergent molecules is a dimerizable or polymerizable functional group selected from the group consisting of thiols, acid hydrazides, aldehydes, amines, and ethylene residues that are suitably substituted to provide enamines upon reaction with an amine. Claim **5** specifies wherein the lipophilic residue is selected from the group consisting of lipophilic amides, esters and ethers. Claim **6** specifies wherein the functional group for binding to nucleic acid molecules is selected from an amine or derivative thereof. Claim **7** specifies wherein the functional group for binding to nucleic acid molecules is guanidine. Claims **8-11** specify wherein the organic cationic precursor molecule is represented by various compositions corresponding to the cited structures named general formula I. Claims **12-21** specify certain side groups for the compositions of general formula I in claims 8-11. Claim **22** specifies wherein the one or more covalent bonds between the cationic molecules are degradable under cellular conditions. Claims **23-26** specify wherein the transfection particle of claim 1 comprises a single nucleic acid molecule, such as a DNA, a DNA plasmid or an RNA. Claim **27** species that the particle of claim 1 is characterized in that it is linked via one or more covalent bonds to one or more cellular targeting functionalities

and/or one or more functionalities capable of facilitating endocytosis. Claim **28** further specifies wherein said functionalities are linked via said one or more covalent bonds to the cationic molecules. Claim **29** specifies wherein said functionalities are linked via said one or more covalent bonds to nucleic acid binding molecules that are present in addition to the cationic molecules. Claim **30** specifies wherein the targeting functionality is a cellular protein ligand. Claims **31-33** further specify wherein the targeting functionality is a sugar residue such as galactose or mannose. Claim **34** specifies that the transfection particle of claim 1 is characterized in that it carries one or more endosomolytic functions. Claim **35** specifies wherein said endosomolytic functions are linked to the cationic molecules. Claim **36** specifies wherein said functions are linked to nucleic acid binding molecules that are present in addition to the cationic molecules. Claim **37** specifies wherein the endosomolytic function is a fusogenic peptide. Claim **38** specifies wherein the endosomolytic function is a virus. Claim **39** specifies wherein the virus is an adenovirus. New claim **48** is drawn to a particle for transfecting higher eukaryotic cells with nucleic acid molecules in vitro or in vivo comprising: (a) one or more nucleic acid molecules; (b) identical or different organic cationic precursor molecules linked to each other via one or more covalent bonds; wherein said precursor molecules are ionically associated with said one or more nucleic acid molecules without forming any crosslinks between said nucleic acid molecules and said cationic precursor molecules, thereby condensing said one or more nucleic acid molecules.

Claim **40** is drawn to a method for preparing transfection particles of claim 1 wherein cationic precursor molecules are added to nucleic acid molecules in a suitable buffer, allowed to form complexes with the nucleic acid and allowed to covalently link to identical or different cationic precursor molecules on the nucleic acid template. Claim **41** specifies wherein the cationic precursor molecules are lipophilic and are allowed to covalently link under mild oxidative conditions.

Claims **42-43** are drawn to pharmaceutical compositions comprising a pharmaceutically effective amount of the transfection particle of claim 1, wherein the nucleic acid molecule is therapeutically active such as a plasmid encoding a therapeutically active protein. Claim **44** is drawn to a method for introducing therapeutically active nucleic acid into a mammal, wherein a transfection particle of claim 1 is administered to said mammal intradermally.

Claims **45-47** are drawn to kits comprising one or more nucleic acid molecules, one or more cationic precursor molecules, one or more cationic precursor molecules, suitable buffers, and other reagents or mechanical devices that are useful for preparation, purification and in vitro or in vivo application of a transfection particle of claim 1; comprising in addition or more functionality for cellular targeting; comprising in addition one or more endosomolytic functionalities.

The claims are drawn to a broad scope of possible compositions for use both in vitro or in vivo. The claims specify pharmaceutical compositions and methods of therapeutic use in mammals. The claims provide several conditions for functionality of the claimed compositions

in addition to therapeutic administration of the nucleic acids (DNA or RNA or DNA plasmids) such as endosomolytic functions linked to the cationic molecules or to the nucleic acid binding molecules. This function relates to the ability of the claimed compositions to function inside the cell after administration of the compositions linked to the therapeutic nucleic acids to either cells in culture or in a whole organism. The above functions are generic to all types of liposomes known in the art for administration of nucleic acids to cells in culture or in whole organisms, but the art teaches selective success of different types of liposomes for delivery of nucleic acids such as gene therapeutic agents. (Such art is discussed below.) The unpredictability in the art for design and use of in vivo therapeutic agents will be discussed below.

The instant claims also have important functional limitations regarding how the claimed liposomal compositions are made. Specifically, claims 40 and 41 teach formation of compositions having a covalent linkages between the cationic precursor molecules and the nucleic acid template where the covalent linkage is allowed to form under mild oxidative conditions. It is this feature of the claimed compositions which is considered to distinguish Applicants' invention from the prior art, since other well-known liposomal compositions are necessarily represented as having covalent linkages between the cationic molecules linked to the nucleic acids. New claim 48 further provides the limitation wherein the cationic precursor molecules are linked to each other via one or more covalent bonds, but the precursor molecules are ionically associated with the nucleic acid molecules. The unpredictability in the art for the

design of the broad scope of claimed compounds in view of such functional considerations in making the claimed compounds is discussed below.

The specification as filed teaches making transfection particles having a di-sulfide bond formed between cysteine residues at the end of an alkyl chain where upon oxidation of the cysteine-alkyl precursor molecules and addition of the DNA template, the stable lipid/DNA particle is formed. The specification exemplifies this in Figure 1, Figure 3, and in the examples. Figure 10 shows that C8-Cys, C11-Cys and C12-Cys were not as effective as C10-Cys. The specification teaches on page 43 that the "oxidation of cysteine detergent $C_{10}C^{G+}$ into cystine lipid $(C_{10}C^{G+})_2$ occurs faster in the presence of template DNA." The specification further teaches by way of example complexing Spermine-N1, N12-bis-cysteineamide and calf-thymus DNA and teaches that "the SC_2 complexes and compacts DNA in the same manner as spermine. On the other hand, the formed particles seemed to be more stable vis a vis the ionic strength of the medium, which is in relation with the oxidation of the thiol functions." (Page 67) Figures 24-26 taught the effect of transferrin-polylysine on the delivery of $(C_{10}C^{G+})_2$/DNA complexes in cells in culture. Example 18 taught intradermal gene delivery of $(C_{10}C^{G+})_2$/ pCMVL DNA complexes in mice but no data on the therapeutic effects of such delivery. Example 20 taught transfection of BNL CL.2 cells with $(C_{12}CO)_2$/DNA complexes (ornithyl-cysteine-dodecylamide detergent as the starting material) for luciferase reporter gene expression.

While the specification as filed is enabling for making and using the exemplified types of liposomes for delivery of nucleic acids to cells in culture and in rodents, the specification is not

enabled for making and using the breadth of claimed compositions for the functions claimed since neither the specification nor the art teach the predictability of substituting any type of precursor molecule with the scope of linkages claimed for administration of any type of nucleic acid especially for therapeutic uses to any whole organism as claimed.

As argued previously in the Official Action mailed 08/08/01, there is a high level of unpredictability for making and using transfection particles for successful achievement of transfection. See paragraph 2 in the Action which cites Zelphati et al. transfection particles known in the art for use in administering therapeutic nucleic acids to cells. While all the claimed limitations are art recognized molecules, their combination to form functional transfection particles does not carry an expectation of success absent specific guidance for the specific structures of precursor molecules, specific substitutions of linkages, etc., concentrations and ratios of concentrations, and the actual steps for formation of the claimed compositions. The closed prior art to the instant Cys-C10 compounds was taught by Staatz et al. (Liebigs Ann. Chem. 1989: (a) pages 51-57 and (b) 127-131). They teach that "in order to synthesize a variable system of well-defined one- and two-chain chiral amphiphils that are able to form liposomes, we choose s-triazine as linking unit between the lipophilic and hydrophilic moieties. The lipophilic part is made of long-chain alcohols or alkylamines, whereas the hydrophilic part of the molecules is formed by the trifunctional amino acids L-cysteine, L-serine, or L-lysine. These are linked to the s-triazine with their w-functional group....On the investigation of their liposome-building properties only the cysteine amphiphiles with two alkyl chains are found to be capable of forming

vesicles." (Abstract of (a)) Although they taught use of s-triazine in making their liposomes, they explicitly taught that the substitution of the precursor molecules did not function to form liposomes. As such, one of skill in the art would not have had an expectation of success to make and use any such transfection particle broadly claimed with any precursor molecule.

Schott et al. (Biochemica et Biophysica Acta 940, 1988, 127-135) further taught the design of palmitoyl derivatives of L-cysteine, cysteamine, L-cystine, cystamine and their incorporation into the bilayers of unilamellar liposomes. They provide specific guidance for the design of liposomes which specifically couple to antibodies but tech several general problems which are unpredictable in the liposome art. For instance on page 128 they teach that "the liposome-antibody complexes are still containing many non-used but activated sulfhydryl residues which are located on the outside as well as on the inner side of the bilayer membrane. When using such liposomes for cell-targeting it can not be excluded that these reactive liposomal groups may cause undesired side reactions. In regard to therapeutical application the possibility of toxical side reactions has to be considered." They further teach that in design of liposomes the "size, homogeneity and stability" of the liposomes must be considered. (Page 128, col. A) On page 133 they taught other considerations of the design of liposomes: "In regard to a therapeutical application of derivatized liposomes we have first of all guaranteed that all components used for derivatization of the liposomes are non-toxic, analytically characterized and easily accessible. Another demand to be met is that the functionalized liposomes are obtained as small unilamellar vesicles in a reproducible population homogeneity (H) and hydrodynamic

diameters (D) and can be stored over a longer period of time." They teach on page 134 the protection of the sulfhydryl residues for the storage of their liposomes in the presence of oxidizing agents to that dimerization won't occur. However, they also teach deprotection by reduction with dithiothreitol if needed. The teach calculation of the number of functional groups per liposome. They teach that "the stability of the liposomes is remarkable as sulfhydryl components not immobilized to liposomes lose their reactivity after some time at neutral pH." they teach practical considerations in the stearic design of such molecules. For example, "liposomes functionalized with phosphatidylethanolamide of carboxyacyl derivatives antibodies could only be coupled if the lipophilic part of the molecule was separated from the carboxylate function by a long spacer." They taught also practical considerations such as cost. "In regard to a therapeutic application (immunotargeting) the efficiency of liposomal loading with expensive antibodies should be as low as possible in order to avoid high costs and prevent unwanted immune reactions....The crucial point for in vivo experiments is that degradation of the biodegradable antibody-liposome complexes in blood and serum proceeds slower than the cell targeting." They thus teach that the composition of the liposome, type of detergent, the concentration of materials, stearic hindrance, ability to form vesicles, number of binding regions, toxicity of the materials, degradation, ability to make (synthesize) a particular chemical design, addition of molecules for cell targeting, etc. are critical features which require experimentation in the art and testing in cells and whole organisms to determine. Due the myriad scope of possibilities in design of any transfecting particle having one or more covalent bonds, for the

claimed functions and intended uses, delivery of nucleic acids, no general guidance is available absent experimentation for the characterization of any such potential compositions.

In regards to the use of any such liposomal composition for transportation of any nucleic acid for the intended uses of such transfection particles in vivo for therapeutic uses, there is a high level of unpredictability in the art for design of not only the transfecting agent, but the type of therapeutic DNA administered. Not every known type of transfection particle is suitable for transfection of any type of nucleic acid molecule. The design of the transfection molecule is thus intimately connected with the type of transfection nucleic acid administered and where the nucleic acid is administered. The instant invention contemplates improved delivery of nucleic acids by design of transfection agents which allow improved endosomolytic release of the nucleic acids, but no specific examples are given demonstrating how such a desired trait is designed into any type of transfection particle as instantly claimed. Furthermore, in regards to administration of any type of transfection particle to a whole organism, not every cell type is accessible equally to administration of such transfection particles. Different cell types thus have different needs and considerations in design and delivery.

Freeman et al. is further cited to teach the considerations necessary in design of liposomes for gene therapy type uses for administration to a whole organism. They thought that at the time the invention was made "in summary, the results highlight some of the issues surrounding the preparation and dosing of DNA to the lungs but demonstrate that transfection can be achieved using DNA in conjunction with the appropriate additive. Along with efforts at the molecular and

cellular level to enhance intracellular translocation of plasmid and increase protein copies, there are means whereby relatively simple formulations, that are readily scalable to manufacturing levels, can also improve transfection. The particular use of bile-salts, opens a new area of investigation and although their use in the lungs may be limited due to toxicity, their use with injectable preparations into tumors for example may be acceptable. More importantly, the results imply that other less toxic derivatives might be developed once a better understanding of the mechanisms of action are obtained." (Page 208) Although the use of bile-salts is not involved in the instant invention, the factors considered unpredictable by Freeman are the same. For instance, (1) routes of administration, (2) toxicity of the therapeutic agent, (3) effect amounts for therapeutic use, (4) understanding the mechanisms of action of the therapeutic compound in the whole organism remain to be highly unpredictable in the art of treatment of a whole organism. These facts directly relate to the design and use of liposomes for carrying therapeutic nucleic acids into whole organisms.

While the instant specification as filed contemplates specifically the design of ($C_{10}C^{G+}$) and ornithyl-cysteine-dodecylamide ($C_{12}CO$) liposomes and methods of making and using said compositions in cells in culture and in rodents, such results do not correlate to making any transfection particle broadly claimed for any use as claimed. One of skill in the art at the time of the invention was made would necessarily had to practice "trial and error" experimentation to make and use the scope of claimed transfection particles since neither the art nor the specification as filed provided sufficient guidance as to how to substitute any of the claimed limitations other

than those exemplified in the specification in a particular concentration and order for the formation of liposomes that stability interact with nucleic acids for delivery into living cells and whole organisms. One of skill in the art would have necessarily practiced undue experimentation to make and use the various claimed compositions absent such guidance.

Response to Arguments

5. Applicant's arguments filed 1/08/02 have been fully considered but they are not persuasive.

The following response addresses the pertinent responses to the previous rejections of claims 8-18 and 42-44.

Applicant points out the legal standard for the enablement requirement on pages 13-14 of the response. Applicant responds as to how Applicants' description enables all the claims, in particular claims 8-18 and 42-44 on pages 15-18. Applicant then argues that the Examiner has not made a prima facie showing of non-enablement and cites several exhibits on pages 18-25.

In view of the above newly recited 35 U.S.C. 112, scope of enablement rejection, it is believed that a prima facie showing of lack of enablement for the breath of the claims is not clearly presented. The Examiners' conclusion was based on the following:

MPEP 2164 teaches the following standards for a determination of whether the specification taught how to make and use the claimed invention at the time the invention was made by weighing whether or not undue experimentation was required to make and use the

invention as claimed. **MPEP 2164.01(a) lists the factors for determining "whether there is sufficient evidence to support a determination that a disclosure does not satisfy the enablement requirement and whether any necessary experimentation is "undue." These factors include, but are not limited to: (A) The breadth of the claims; (B) The nature of the invention; C) The state of the prior art; (D) The level of one of ordinary skill; (E) The level of predictability in the art; (F) the amount of direction provided by the inventor; (G) The existence of working examples; and (H) The quantity of experimentation needed to make or use the invention based on the content of the disclosure.** ***In re Wands*****, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988)"**

In view of a review of the liposome art at the time the invention was made, especially a review of the art directed to use of SH bonds and oxidation as a mechanism for formation of the liposome composition, it was determined that the quantity of experimentation needed to make and use the scope of the claimed constructs was undue. Specific references were cited which taught the unpredictable factors in making and using related liposomal compositions. It is thus believed that the legal standard for determination of enablement of the claimed invention was met in the above analysis of instant claims 1-48 and that the disclosure was not commensurate with the scope of protection sought by the claims for enabling one of skill in the art to make any cationic precursor without crosslinking for the formation of functional transfecting particles.

Applicant argues that to be fully enabled the description only needs to enable a person of ordinary skill in the art in making the claimed transfection particles and in using them in at least

one manner. In response, while to show a general utility of an invention (ie. to meet the utility requirements under 35 U.S.C. 101) only one use is necessary, the standards for the coupled ability to make and use a claimed compound under 35 U.S.C. 112, require a total consideration of all the above listed factors. Thus, the standard is not based on finding at least one use for the claimed compounds, but determining whether the entire breadth of the claimed compounds is enabled by the art and disclosure. In the instant case the claims are drawn to any cationic precursor particle condensed without crosslinking. The ability to synthesize any such molecule so that it would have an expectation to have a particular use is not clearly represented in the art or the disclosure for the specific reasons argued above. Since the disclosed embodiments are not representative of the entire scope of claimed compositions, one of skill in the art would not have been given sufficient guidance to make and use any such composition broadly claimed. Thus, although there is one functional composition taught in the specification, this is not representative of the breadth of possible compositions instantly claimed.

Applicant argues on pages 15 and 16 of the response that the specification provides description of methods of making such transfection particles and guidance including how to calculate the ratio of cationic precursor to nucleotide molecules, how long and at what temperature to complex the cationic precursors to the nucleotide molecules, under what conditions to affect oligomerization between cationic precursors, and how to monitor the formation of complexation. Applicant also argues that guidance as to the cationic precursor molecules used to make the transfection particles is given.

In response, the specification very generally provides such guidance which is not sufficient for the breadth of claimed compositions considered by the instant claims. The type of guidance provided requires one of skill in the art to perform "trial and error"experimentation to overcome the unpredictable factors cited above. Although there are many known tools in the art for measuring the formation of liposomal compositions, knowledge of such tools does not provide the necessary steps for determination of which of the broad scope of claimed starting materials will work together to form the claimed types of liposomal compositions claimed.

Applicant further argues that the description further taught how to transfect the claimed compositions without undue experimentation. In response, it is agreed that the method of transfecting cells in cell culture is not a serious burden on one of skill in the art. Although the exact concentration of liposomes must be determined so that the transfection is not toxic, such assays are easily performed by use of concentration gradients in the laboratory. However, use of the claimed compounds in vivo has a much higher level of unpredictability in the art since the complexity of the invention is much higher. On pages 17 and 18 Applicant argues that several enabling in vivo uses are described and that the skilled artisan would know how to formulate a pharmaceutical composition which can be administered, for example, intradermally. However, as argued above, there is not a correlation in the art between administration of one therapeutic agent and another in a whole organism. The number of variable factors to be considered in therapeutic administration of compositions is much higher than the number of factors to be considered in administration of a compound to cells in culture. Several references are cited

above to teach that overcoming such issues are not routine in the art. The pharmaceutical formulation which functions for one type of therapeutic molecule does not correlate to the formulation of other types of compounds when the structures of such molecules differ substantially. In the instant case, neither the art nor the specification taught specific formulation for the instant compounds for an effective desired therapeutic effect. Although, the specification teaches one type of administration of one type of molecule, such data does not provide specific guidance for administration of any liposome/DNA complexes by any route of administration for any in vivo use as broadly claimed. The guidance in Examples 16-19 in the instant specification does not provide guidance for any type of liposomal composition administered to any whole organism without "trial and error" experimentation to ascertain specific concentrations, formulations, etc.

Applicant argues on the bottom of page 18 that "a patent applicant's specification disclosure which contains a teaching of how to make and use the invention must be taken as enabling unless the Patent Office provides sufficient reason to doubt the accuracy of the disclosure.... Here, Applicants submit that the Examiner has provided no evidence that a skilled artisan would doubt the enablement of the claimed polynucleotide. The Examiner has not met her burden in explaining why the skilled artisan would not be enabled to practice the claimed invention throughout the entire scope of the claims."

In response, while the above 35 U.S.C. 112, scope of enablement, rejection does not doubt the "accuracy of the disclosure", it does persist with the argument that neither the

disclosure as filed nor the art at the time the invention was made provided sufficient guidance to one of skill in the art to make and use the scope of the claimed liposomal compositions. The specific arguments to the basis of the rejection made in the previous Action (pages 19-23 in the instant response) are not specifically considered in view of the revised rejection made above. However, the exhibits provided with the response are addressed below.

Applicants' quote several review references describing the state of the liposome art at the time the invention was made: Maurer et al., Schatzlein and Felgner et al. While these references do provide a nice overview of the state of the art at the time the invention was made, they mainly provide discussion of well-known liposomes such as DOTMA and DOPE (Mahato et al. Page 853) and others (page 854 of Mahato et al.) which as Applicant pointed out in the response do not form covalent linkages with the nucleic acids. Applicant's quote Maurer et al.: "[c]ationic liposomes are the most widely and successfully used lipid-based vectors for gene transfer.... The preparation procedure is simple. The cationic liposomes...are mixed with DNA in a dilute solution. The complexes form spontaneously due to electrostatic charge interactions, which lead to liposome fusion and aggregation." The fact that well-known cationic liposomes form electrostatic charge interactions with different DNA molecules does not necessarily apply to the instantly claimed invention with novel combinations of cationic precursor molecules. Furthermore, in regards to the first full paragraph on page 24 of the response, it is not disputed that liposomes are well-known for in vitro transfection. Finally, Applicants' cite Felgner et al. , page 134-135, to teach that the skilled artisan at the time of Applicants' invention was familiar

with therapeutic in vivo uses of cationic lipids. However, Felgner et al. discusses use of known lipids with viral vectors for delivery of gene therapeutic agents. Such compositions do not correlated to the novel combinations of cationic molecules instantly claimed and formulated in novel ways. Felgner et al. further supports the arguments made above that there is still unpredictability in the art for the administration of liposomes in vivo in regards to non-specific binding. They also teach that enhancement is needed for controlled DNA condensation and product monodispersion systems, escape from the endosomal compartment, and concentration of the nucleic acids to the nucleus. While the instant specification considers such issues prophetically in relation to the disclosed liposomes, the specification does not provide sufficient guidance that the claimed compositions would have an expectation to display such improvements absent "trial and error" experimentation.

In summary, while the references cited do display a picture of the successes and pitfalls of liposomal use at the time the invention was made, they do not further support the deficiencies in the instance specification as filed to provide adequate guidance for making and using the scope of claimed compounds.

6. The claims are considered free of the prior art since the closest prior art is discussed above and does not specifically read on making cationic precursors with covalent bonds without crosslinking the lipid.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to *Mary M. Schmidt*, whose telephone number is (703) 308-4471.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, *John LeGuyader*, may be reached at (703) 308-0447.

Any inquiry of a general nature or relating to the status of this application should be directed to the Group Analyst, *Katrina Turner*, whose telephone number is (703) 305-3413.

JOHN L. LeGUYADER
SUPERVISORY PATENT EXAMINER
TECHNOLOGY CENTER 1600

M. M. Schmidt
March 23, 2002